Title	Allergen related gene expression in apple fruit is differentially controlled by ethylene
	during ripening
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Abstract

Certain proteins or families of proteins present in plants and in many fruits can act as allergens that may cause an overreaction of the human immune system. Recent findings showed that apple consumption can cause allergic reactions in some consumers, especially among northern and central European populations, due to the presence of allergenic proteins. Investigations of apple fruit allergens have indicated that production of allergens is influenced by many biotic and abiotic factors. To better understand the regulation of allergen production during fruit ripening, and to examine the influence of ethylene on expression of genes encoding allergens in fruit, apples harvested at the pre-climacteric stage were allowed to ripen naturally, or ripening was either stimulated by treatment with 36 μ L L⁻¹ ethylene for 24 h or inhibited with 1-MCP treatment (1 μ L L⁻¹ for 24 h). As an indicator of physiological status, ethylene production, was monitored up to 21 d after ethylene treatment or up to 47 d after 1-MCP treatment. Realtime qPCR was used to identify allergen genes that were differentially expressed in ethylene- and 1-MCPtreated fruit. Sixteen allergen genes representing four gene families were investigated. Transcript abundance of several genes was found to change significantly during ripening. Genes encoding Mal d1.01, Mal d1b, Mal dld, Mal dle, Mal dl-associated protein (MdAP), and Mal d4.01 were significantly up-regulated in fruit during ripening, and further enhanced after ethylene treatment. By contrast, expression of Mal d1.04, Mal d2.01, Mal d2.02, Mal d2.03, Mal d3.01, and Mal d4.03 genes decreased. Declining expression of Mal d3.01, Mal d4.02, and Mal d4.03 during fruit ripening was also found. Treatment with 1-MCP and ethylene generally produced opposite effects, which provides additional evidence that regulation of these genes is ethylene dependent. Overall, changes in the transcriptional profiles of genes encoding apple fruit allergens during ripening and senescence, and in response to ethylene, were complex and dynamic. Characterization of changes in these allergens during storage will potentially aid in the control of quality and safety of apple fruit.